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(FILE 'HOME' ENTERED AT 16:00:34 ON 19 SEP 2002)

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BIOBUSINESS, BIOCOMMERCE, BIOSIS, BIOTECHABS, BIOTECHDS, BIOTECHNO,
CABA,
CANCERLIT, CAPLUS, CEABA-VTB, CEN, CIN, CONFSCI, CROPB, CROPU, DDFB,
DDFU, DGENE, DRUGB, DRUGLAUNCH, DRUGMONOG2, ...' ENTERED AT 16:00:41 ON
19 SEP 2002

SEA (FUSION PROTEIN OR HYBRID PROTEIN OR BI-FUNCTIONAL
PROTEIN)

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L1 QUE (FUSION PROTEIN OR HYBRID PROTEIN OR BI-FUNCTIONAL
PROTEIN)

FILE 'MEDLINE, BIOSIS, CAPLUS, EMBASE, BIOTECHNO, SCISEARCH' ENTERED AT
16:04:56 ON 19 SEP 2002

L2 422 S L1 (S) (CELLULOSE BINDING DOMAIN OR CBD)
L3 43 S L2 (S) ANTIBODY
L4 12 DUP REM L3 (31 DUPLICATES REMOVED)

=> d 14 ibib ab 1-12

L4 ANSWER 1 OF 12 CAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 2002:355978 CAPLUS
DOCUMENT NUMBER: 137:105888
TITLE: Use of recombinant cellulose-binding domains of
Trichoderma reesei cellulase as a selective
immunocytochemical marker for cellulose in protozoa
AUTHOR(S): Linder, Markus; Winiecka-Krusnell, Jadwiga; Linder,
Ewert
CORPORATE SOURCE: VTT Biotechnology, Espoo, FIN 02044-VTT, Finland
SOURCE: Applied and Environmental Microbiology (2002), 68(5),
2503-2508
CODEN: AEMIDF; ISSN: 0099-2240
PUBLISHER: American Society for Microbiology
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Some unicellular organisms are able to encyst as a protective response to
harmful environment. The cyst wall usually contains chitin as its main
structural constituent, but in some cases, as in Acanthamoeba, it
consists
of cellulose instead. Specific cytochem. differentiation between
cellulose and chitin by microscopy has not been possible, due to the
similarity of their constituent .beta.-1,4-linked hexose backbones.
Thus,
various fluorescent brightening agents and lectins bind to both cellulose
and chitin. We have used a recombinant cellulose-binding protein
consisting of two cellulose-binding domains (CBDs) from Trichoderma
reesei
cellulases linked together in combination with monoclonal anticellulase
antibodies and anti-mouse Ig fluorescein conjugate to specifically stain
cellulose in the cysts of Acanthamoeba strains for fluorescence
microscopy
imaging. Staining was obsd. in ruptured cysts and frozen sections of
cysts but not in intact mature cysts. No staining reaction was obsd.
with
the chitin-contg. cyst walls of Giardia intestinalis, Entamoeba dispar,
or
Pneumocystis carinii. Thus, the recombinant CBD can be used as a marker
to distinguish between cellulose and chitin. Thirteen of 25
environmental
or clin. isolates of amoebae reacted in the CBD binding assay. All 13
isolates were identified as Acanthamoeba spp. Five isolates of
Hartmannella and seven isolates of Naegleria tested neg. in the CBD
binding assay. Whether cyst wall cellulose really is a unique property
of
Acanthamoeba spp. among free-living amoebae, as suggested our findings,
remains to be shown in more extensive studies.

REFERENCE COUNT: 39 THERE ARE 39 CITED REFERENCES AVAILABLE FOR
THIS

RECORD. ALL CITATIONS AVAILABLE IN THE RE
FORMAT

L4 ANSWER 2 OF 12 CAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 2001:472868 CAPLUS
DOCUMENT NUMBER: 135:78577
TITLE: Method of delivering benefit agent to fabric via
antibody/fusion protein as binding molecule
INVENTOR(S): Howell, Steven; Little, Julie; Van Der Logt, Cornelis

Paul Erik; Parry, Neil James
 PATENT ASSIGNEE(S): Unilever N.V., Neth.; Unilever Plc; Hindustan Lever Ltd
 SOURCE: PCT Int. Appl., 69 pp.
 CODEN: PIXXD2
 DOCUMENT TYPE: Patent
 LANGUAGE: English
 FAMILY ACC. NUM. COUNT: 1
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2001046356	A2	20010628	WO 2000-EP12529	20001208
WO 2001046356	A3	20020110		
W: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CR, CU, CZ, DE, DK, DM, DZ, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM RW: GH, GM, KE, LS, MW, MZ, SD, SL, SZ, TZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, TR, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG US 2002019324 A1 20020214 US 2000-742693 20001220				

PRIORITY APPLN. INFO.: EP 1999-310431 A 19991222
 AB A method of delivering a benefit agent to fabric for exerting a pre-detd. activity useful for stain removal, perfume delivery, and treating collars and cuffs for wear, is provided, wherein the fabric is pre-treated with a multi-specific binding mol. which has a high binding affinity to said fabric through one specificity and is capable of binding to said benefit agent through another specificity, followed by contacting said pre-treated fabric with said benefit agent, to enhance said pre-detd. activity to said fabric. Preferably, the binding mol. is an **antibody** or fragment thereof, or a **fusion protein** comprising a **cellulose binding domain** and a domain having a high binding affinity to another ligand which is directed to said benefit agent such as glucose oxidase.

L4 ANSWER 3 OF 12 MEDLINE DUPLICATE 1
 ACCESSION NUMBER: 2000275530 MEDLINE
 DOCUMENT NUMBER: 20275530 PubMed ID: 10814589
 TITLE: Expression, purification and applications of staphylococcal protein A fused to cellulose-binding domain.
 AUTHOR: Shpigel E; Goldlust A; Eshel A; Ber I K; Efroni G; Singer Y; Levy I; Dekel M; Shoseyov O
 CORPORATE SOURCE: The Kennedy Leigh Centre for Horticulture Research and The Otto Warburg Center for Agricultural Biotechnology, The Faculty of Agricultural, Food and Environmental Quality Sciences, The Hebrew University of Jerusalem, P.O. Box 12, Rehovot, Israel.
 SOURCE: BIOTECHNOLOGY AND APPLIED BIOCHEMISTRY, (2000 Jun) 31 (Pt 3) 197-203.
 Journal code: 8609465. ISSN: 0885-4513.
 PUB. COUNTRY: ENGLAND: United Kingdom
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 200007
 ENTRY DATE: Entered STN: 20000810
 Last Updated on STN: 20000810
 Entered Medline: 20000727

AB Because staphylococcal Protein A (ProtA) binds specifically to IgG, it has

been used for many immunological manipulations, most notably **antibody** purification and diagnostics. Immobilization is required for most of these applications. Here we describe a genetic-engineering approach to immobilizing ProtA on cellulose, by fusing it to **cellulose-binding domain (CBD)** derived from the cellulose-binding Protein A of *Clostridium cellulovorans*.

The bifunctional **fusion protein** was expressed in *Escherichia coli*, recovered on a cellulose column and purified by elution at alkaline pH. ProtA-CBD was used to purify IgG from rabbit serum and its ability to bind IgG from different sources was determined. The bifunctional chimaeric protein can bind up to 23.4 mg/ml human IgG at a ratio of 1 mol of ProtA-CBD/2 mol of human IgG, and can purify up to 11.6 mg/ml rabbit IgG from a serum. The ability to bind functionally active CBD-affinity reagents to cellulosic microtitre plates was demonstrated. Our results indicate that a combination of CBD-affinity reagents and cellulosic microtitre plates is an attractive diagnostics matrix for the following reasons: (i) cellulose exhibits very low non-specific binding; and (ii) **CBD-fusion proteins** bind directly to cellulose at high density. A unique signal-amplification method was developed based on the ability of ProtA-CBD to link stained cellulose particles to primary **antibody** in a Western blot.

L4 ANSWER 4 OF 12 CAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 2000:779465 CAPLUS
DOCUMENT NUMBER: 134:38967
TITLE: Phage display of cellulose binding domains for biotechnological application
AUTHOR(S): Benhar, Itai; Tamarkin, Aviva; Marash, Lea; Berdichevsky, Yevgeny; Yaron, Sima; Shoham, Yuval; Lamed, Raphael; Bayer, Edward A.
CORPORATE SOURCE: Department of Molecular Microbiology and Biotechnology, The George S. Wise Faculty of Life Sciences, Tel-Aviv University, Ramat Aviv, Israel
SOURCE: ACS Symposium Series (2000), 769 (Glycosyl Hydrolases for Biomass Conversion), 168-189
CODEN: ACSMC8; ISSN: 0097-6156
PUBLISHER: American Chemical Society
DOCUMENT TYPE: Journal; General Review
LANGUAGE: English

AB A review with 65 refs. In recent years, cellulose-binding domains (CBDs) derived from the cellulolytic systems of cellulose-degrading microorganisms have become a focal point of attention for a wide range of biotechnol. applications. The low cost and availability of cellulose matrixes have rendered CBDs attractive as affinity tags for the purification and immobilization of a plethora of proteins. Intensive studies of cellulose degradation pathways and the identification of components of the cellulose-degrading machinery have contributed significantly to our understanding of the structure and function of CBDs. The time is now ripe to utilize engineered CBDs to improve existing applications and to devise novel ones. Here we describe our recent results of experiments where the *Clostridium thermocellum* scaffoldin CBD was genetically engineered for such purposes. We describe the development of a novel phage display system where the *C. thermocellum* **CBD** is displayed as a **fusion protein** with single-chain **antibodies**. Our system is a filamentous (M13) phage display system that enables the efficient isolation and.

REFERENCE COUNT: 65 THERE ARE 65 CITED REFERENCES AVAILABLE FOR THIS

FORMAT RECORD. ALL CITATIONS AVAILABLE IN THE RE

L4 ANSWER 5 OF 12 CAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 1999:530317 CAPLUS
 DOCUMENT NUMBER: 131:181955
 TITLE: Purification of recombinantly prepared proteins by using the cellulose-binding domain of a cellulose-degrading enzyme as an affinity tag
 INVENTOR(S): Karita, Shuichi; Ohmiya, Kunio; Sakka, Kazuo; Kimura, Tetsuya
 PATENT ASSIGNEE(S): Toyobo Co., Ltd., Japan
 SOURCE: Jpn. Kokai Tokkyo Koho, 12 pp.
 CODEN: JKXXAF
 DOCUMENT TYPE: Patent
 LANGUAGE: Japanese
 FAMILY ACC. NUM. COUNT: 1
 PATENT INFORMATION:

	PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
	JP 11225763	A2	19990824	JP 1998-29410	19980212

AB Purifn. of a protein (enzyme, **antibody**, or hormone) that is expressed as a **fusion protein** with the **cellulose-binding domain (CBD)** of a cellulose-degrading enzyme is described. The fusion protein-contg. cellular ext. is first mixed with an insol., non-crystal cellulose carrier for absorption; the absorbed fusion protein is then eluted with a carbohydrate such as cellobiose, maltose, glucose, or xylose. The target protein is then retrieved from the purifd. fusion protein by digestion with a proteinase such as trypsin. Purifn. of endoglucanase IV of Ruminococcus albus fused with the CBD of xylanase A of Clostridium stercorarium was described.

L4 ANSWER 6 OF 12 MEDLINE DUPLICATE 2
 ACCESSION NUMBER: 1999160408 MEDLINE
 DOCUMENT NUMBER: 99160408 PubMed ID: 10049766
 TITLE: A cellulose-binding domain-fused recombinant human T cell connective tissue-activating peptide-III manifests heparanase activity.
 AUTHOR: Rechter M; Lider O; Cahalon L; Baharav E; Dekel M; Seigel D; Vlodavsky I; Aingorn H; Cohen I R; Shoseyov O
 CORPORATE SOURCE: Department of Immunology, The Weizmann Institute of Science, Rehovot, 76100, Israel.
 SOURCE: BIOCHEMICAL AND BIOPHYSICAL RESEARCH COMMUNICATIONS, (1999 Feb 24) 255 (3) 657-62.
 Journal code: 0372516. ISSN: 0006-291X.
 PUB. COUNTRY: United States
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 199904
 ENTRY DATE: Entered STN: 19990413
 Last Updated on STN: 19990413
 Entered Medline: 19990401

AB The chemokine connective tissue-activating peptide (CTAP)-III, which belongs to the leukocyte-derived growth factor family of mediators, was previously shown to be mitogenic for fibroblasts. However, it has recently been shown that CTAP-III, released from platelets, can act like a heparanase enzyme and degrade heparan sulfate. This suggests that CTAP-III may also function as a proinflammatory mediator. We have successfully cloned CTAP-III from a lambdagt11 cDNA library of PHA-activated human CD4(+) T cells and produced recombinant CTAP-III as a **fusion protein** with a **cellulose-binding domain** moiety. This recombinant CTAP-III exhibited heparanase activity and released degradation products from metabolically labeled, naturally produced extracellular matrix. We have also developed polyclonal

and monoclonal **antibodies**, and these **antibodies**
against the recombinant CTAP-III detected the CTAP-III molecule in human

T

cells, polymorphonuclear leukocytes, and placental extracts. Thus, our study provides tools to examine further immune cell behavior in inflamed sites rich with extracellular moieties and proinflammatory mediators.
Copyright 1999 Academic Press.

L4 ANSWER 7 OF 12 MEDLINE DUPLICATE 3
ACCESSION NUMBER: 2000014693 MEDLINE
DOCUMENT NUMBER: 20014693 PubMed ID: 10545273
TITLE: Matrix-assisted refolding of single-chain Fv- cellulose binding domain fusion proteins.
AUTHOR: Berdichevsky Y; Lamed R; Frenkel D; Gophna U; Bayer E A; Yaron S; Shoham Y; Benhar I
CORPORATE SOURCE: Department of Molecular Microbiology and Biotechnology, The George S. Wise Faculty of Life Sciences, Tel-Aviv University, Ramat Aviv, 69978, Israel.
SOURCE: PROTEIN EXPRESSION AND PURIFICATION, (1999 Nov) 17 (2) 249-59.
Journal code: 9101496. ISSN: 1046-5928.
PUB. COUNTRY: United States
DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals
ENTRY MONTH: 200001
ENTRY DATE: Entered STN: 20000124
Last Updated on STN: 20000124
Entered Medline: 20000112

AB We describe a method for the isolation of recombinant single-chain **antibodies** in a biologically active form. The single-chain **antibodies** are fused to a **cellulose binding domain** as a single-chain protein that accumulates as insoluble inclusion bodies upon expression in Escherichia coli. The inclusion bodies are then solubilized and denatured by an appropriate chaotropic solvent, then reversibly immobilized onto a cellulose matrix via specific interaction of the matrix with the **cellulose binding domain** (CBD) moiety. The efficient immobilization that minimizes the contact between folding protein molecules, thus preventing their aggregation, is facilitated by the robustness of the Clostridium thermocellum CBD we use. This CBD is unique in retaining its specific cellulose binding capability when solubilized in up to 6 M urea, while the proteins fused to it are fully denatured. Refolding of the **fusion proteins** is induced by reducing with time the concentration of the denaturing solvent while in contact with the cellulose matrix. The refolded single-chain **antibodies** in their native state are then recovered by releasing them from the cellulose matrix in high yield of 60% or better, which is threefold or higher than the yield obtained by using published refolding protocols to recover the same scFvs. The described method should have general applicability for the production of many protein-CBD fusions in which the fusion partner is insoluble upon expression.
Copyright 1999 Academic Press.

L4 ANSWER 8 OF 12 MEDLINE DUPLICATE 4
ACCESSION NUMBER: 2000027007 MEDLINE
DOCUMENT NUMBER: 20027007 PubMed ID: 10556552
TITLE: Phage display of a cellulose binding domain from Clostridium thermocellum and its application as a tool for antibody engineering.

AUTHOR: Berdichevsky Y; Ben-Zeev E; Lamed R; Benhar I
 CORPORATE SOURCE: Department of Molecular Microbiology, The George S. Wise
 Faculty of Life Sciences, Green Building, Room 202,
 Tel-Aviv University, Ramat Aviv 69978, Israel.
 SOURCE: JOURNAL OF IMMUNOLOGICAL METHODS, (1999 Aug 31) 228 (1-2)
 151-62.
 Journal code: 1305440. ISSN: 0022-1759.
 PUB. COUNTRY: Netherlands
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 199912
 ENTRY DATE: Entered STN: 20000113
 Last Updated on STN: 20000113
 Entered Medline: 19991228

AB Phage display of **antibody** fragments has proved to be a powerful
 tool for the isolation and in vitro evolution of these biologically
 important molecules. However, the general usefulness of this technology
 is still limited by some technical difficulties. One of the most
 debilitating obstacles to the widespread application of the technology is the
 accumulation of "insert loss" clones in the libraries; phagemid clones
 from which the DNA encoding part or all of the cloned **antibody**
 fragment had been deleted. Another difficulty arises when phage
 technology is applied for cloning hybridoma-derived **antibody** genes, where
 myeloma derived light chains, irrelevant to the hybridoma's
antibody specificity may be fortuitously cloned. Here, we report
 the construction of a novel phage-display system designed to address
 these problems. In our system a single-chain Fv (scFv) is expressed as an
 in-frame **fusion protein** with a **cellulose-**
binding domain (CBD) derived from the
 Clostridium thermocellum cellulosome. The **CBD** domain serves as
 an affinity tag allowing rapid phage capture and concentration from crude
 culture supernatants, and immunological detection of both displaying
 phage and soluble scFv produced thereof. We demonstrate the utility of our
 system in solving the technical difficulties described above, and in
 speeding up the process of scFv isolation from combinatorial
antibody repertoires.

L4 ANSWER 9 OF 12 MEDLINE DUPLICATE 5
 ACCESSION NUMBER: 1999201269 MEDLINE
 DOCUMENT NUMBER: 99201269 PubMed ID: 10099473
 TITLE: Improved immobilization of fusion proteins via
 cellulose-binding domains.
 AUTHOR: Linder M; Nevanen T; Soderholm L; Bengs O; Teeri T T
 CORPORATE SOURCE: VTT Biotechnology and Food Research, P.O. Box 1500,
 FIN-02044, Finland.
 SOURCE: BIOTECHNOLOGY AND BIOENGINEERING, (1998 Dec 5) 60 (5)
 642-7.
 Journal code: 7502021. ISSN: 0006-3592.
 PUB. COUNTRY: United States
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 199904
 ENTRY DATE: Entered STN: 19990511
 Last Updated on STN: 19990511
 Entered Medline: 19990426

AB **Cellulose-binding domains (CBDs)**
 are structurally and functionally independent, noncatalytic modules found
 in many cellulose or hemicellulose degrading enzymes. Recent
 biotechnological applications of the **CBDs** include facilitated

protein immobilization on cellulose supports. In some occasions there have been concerns about the stability of the **CBD** driven immobilization. Here we have studied the chromatographic behavior of variants of the *Trichoderma reesei* cellobiohydrolase I **CBD** belonging to family I. Both **CBDs** fused to **antibody** fragments and isolated **CBDs** were studied and compared. Tritium labeling by reductive methylation was used as a sensitive detection method. The **fusion protein** as well as the isolated **CBD** was found to leak from the column at a rate of 0.3-0.5% of the immobilized protein per column volume. However, the leakage could be overcome by using two **CBDs** instead of a single **CBD** for the immobilization. In this way leakage was reduced to less than 0.01% per column volume. The improved immobilization could also be seen as a decreased migration of the protein down the column in extended washes. Copyright 1998 John Wiley & Sons, Inc.

L4 ANSWER 10 OF 12 BIOSIS COPYRIGHT 2002 BIOLOGICAL ABSTRACTS
INC.DUPLICATE

6

ACCESSION NUMBER: 1997:109020 BIOSIS
DOCUMENT NUMBER: PREV199799408223
TITLE: Comparison of the adsorption properties of a single-chain antibody fragment fused to a fungal or bacterial cellulose-binding domain.
AUTHOR(S): Reinikainen, Tapani; Takkinen, Kristiina; Teeri, Tuula T. (1)
CORPORATE SOURCE: (1) VTT Biotechnol. Food Res., P.O. Box 1500, FIN-02044 VTT
Finland
SOURCE: Enzyme and Microbial Technology, (1997) Vol. 20, No. 2, pp. 143-149.
ISSN: 0141-0229.
DOCUMENT TYPE: Article
LANGUAGE: English

AB *Trichoderma reesei* cellobiohydrolase I (CBHI) and *Cellulomonas fimi* cellulase-xylanase (Cex) both have distinct C-terminal **cellulose-binding domains** which belong to different **CBD** sequence families. Two **fusion proteins** comprising a single-chain **antibody** fragment (OxscFv) against 2-phenyloxazolone fused to the two **CBDs** (**CBD**-CBHI or **CBD**-Cex) were constructed. The binding properties of the **fusion proteins** were studied on different cellulosic substrates. It was shown that the **CBD**-Cex binds the **fusion protein** to cellulose more effectively than the **CBD**-CBHI; however, once immobilized, both **fusion proteins** could be eluted from cellulose only with denaturing agents or very low or high pH. Both **fusion proteins** retained equally well their ability to bind the hapten recognized by their **antibody** part.

L4 ANSWER 11 OF 12 MEDLINE DUPLICATE 7
ACCESSION NUMBER: 97077619 MEDLINE
DOCUMENT NUMBER: 97077619 PubMed ID: 8920186
TITLE: Characterization of *Escherichia coli* expressing an Lpp'OmpA(46-159)-PhoA fusion protein localized in the outer membrane.
AUTHOR: Stathopoulos C; Georgiou G; Earhart C F
CORPORATE SOURCE: Department of Microbiology, University of Texas, Austin, 78712, USA.
SOURCE: APPLIED MICROBIOLOGY AND BIOTECHNOLOGY, (1996 Mar) 45 (1-2)

112-9.
 Journal code: 8406612. ISSN: 0175-7591.
 PUB. COUNTRY: GERMANY: Germany, Federal Republic of
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 LANGUAGE: English
 FILE SEGMENT: Biotechnology
 ENTRY MONTH: 199701
 ENTRY DATE: Entered STN: 19970128
 Last Updated on STN: 19970128
 Entered Medline: 19970102

AB The Lpp'OmpA(46-159) **hybrid protein** can serve as an efficient targeting vehicle for localizing a variety of procaryotic and eucaryotic soluble proteins onto the E. coli surface, thus providing a system for several possible biotechnology applications. Here we show that fusion between Lpp'OmpA(46-159) and bacterial alkaline phosphatase (PhoA), a normally periplasmic dimeric enzyme, are also targeted to the outer membrane. However, protease accessibility experiments and immunoelectron microscopy revealed that, unlike other periplasmic proteins, the PhoA domain of these fusions is not exposed on the cell surface in cells having an intact outer membrane. Conditions that affect the formation of disulfide bonds and the folding of the PhoA domain in the periplasm not only did not facilitate targeting to the cell surface but led to lethality when the fusion was expressed from a high-copy-number plasmid. Furthermore, E. coli expressing the Lpp'OmpA(46-159)-PhoA fusion exhibited strain- and temperature-dependent alterations in outer-membrane permeability. Our results are consistent with previous studies with other vehicles indicating that PhoA is not displayed on the surface when fused to cell-surface expression vectors. Presumably, the enzyme rapidly assumes a tightly folded dimeric conformation that cannot be transported across the outer membrane. The large size and quaternary structure of PhoA may define a limitation of the Lpp'OmpA(46-159) fusion system for the display of periplasmic proteins on the cell surface. Alkaline phosphatase is a unique protein among a group of five periplasmic proteins (beta-lactamase, alkaline phosphatase, Cex cellulase Cex **cellulose-binding domain**, and a single-chain Fv **antibody** fragment), which have been tested as passengers for the Lpp'OmpA(46-159) expression system to date, since it was the only protein not displayed on the surface.

L4 ANSWER 12 OF 12 MEDLINE DUPLICATE 8
 ACCESSION NUMBER: 95133949 MEDLINE
 DOCUMENT NUMBER: 95133949 PubMed ID: 7832524
 TITLE: The expression of recombinant proteins on the external surface of Escherichia coli. Biotechnological applications.
 AUTHOR: Francisco J A; Georgiou G
 CORPORATE SOURCE: Department of Chemical Engineering, University of Texas at Austin 78712.
 SOURCE: ANNALS OF THE NEW YORK ACADEMY OF SCIENCES, (1994 Nov 30) 745 372-82. Ref: 37
 Journal code: 7506858. ISSN: 0077-8923.
 PUB. COUNTRY: United States
 DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
 General Review; (REVIEW)
 (REVIEW, TUTORIAL)
 LANGUAGE: English
 FILE SEGMENT: Priority Journals
 ENTRY MONTH: 199502
 ENTRY DATE: Entered STN: 19950307
 Last Updated on STN: 19950307

AB The expression of recombinant proteins on the external surface of Gram-negative bacteria is expected to open the way for a number of significant biotechnological applications, including the development of live bacterial vaccines, the production of whole cell adsorbents, the preparation of whole cell catalysts, and the display and selection of peptide and **antibody** libraries. We have developed a **fusion protein** system for the production of active recombinant proteins on the surface of Escherichia coli. Using this system we have expressed beta-lactamase, the Cellulomonas fimi exoglucanase Cex as well as its **cellulose binding domain**, and an antidigoxin single chain Fv **antibody** fragment on the cell surface. Recently we have begun to explore some of the potential applications for cell-surface expression.

L6 ANSWER 4 OF 23 CAPLUS COPYRIGHT 2003 ACS

ACCESSION NUMBER: 2000:14964 CAPLUS

DOCUMENT NUMBER: 132:83390

TITLE: Topical cosmetic, dermatological, hygienic, or pharmaceutical composition containing antibodies

INVENTOR(S): Breton, Lionel; Pineau, Nathalie; Giacomoni, Paolo

PATENT ASSIGNEE(S): L'Oreal, Fr.

SOURCE: PCT Int. Appl., 40 pp.

CODEN: PIXXD2

DOCUMENT TYPE: Patent

LANGUAGE: French

FAMILY ACC. NUM. COUNT: 1

PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 2000000163	A1	20000106	WO 1999-FR1549	19990628

W: JP, KR, US

RW: AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE

FR 2780286	A1	19991231	FR 1998-8341	19980630
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PRIORITY APPLN. INFO.: FR 1998-8341 19980630

AB The invention concerns compns. for topical use, comprising a cosmetic, dermatol., hygienic or pharmaceutical medium, characterized in that it further comprises: at least a first antibody contg. a protein domain recognizing a skin and/or nail and/or lip and/or eyelash epitope, said first antibody being coupled or not with a pigment and/or coloring agent and/or active cosmetic or dermatol. principle; at least a second antibody contg. a protein domain recognizing said first antibody epitope and/or a second antibody contg. a protein domain recognizing a pigment and/or a coloring agent and/or an active cosmetic or dermatol. principle, said second antibodies being coupled or not with a pigment and/or coloring agent and/or an active cosmetic or dermatol. principle. Monoclonal and polyclonal antibodies were prepd. and coupled with a coloring agents. A cosmetic powder contained non-coupled anti(yellow iron oxide) **antibody** 1.6, non-coupled anti(red iron oxide) **antibody** 1.7, non-coupled anti(black iron oxide) **antibody** 1.5, yellow iron oxide 1.6, red iron oxide 1.7, black iron oxide 1.5, Me paraben 0.3, **perfume** 0.2, magnesium carbonate 0.2, talc 73.8, titanium oxide, alumina, glycerin and silica 3, nylon-12 10, dimethicone 2.51, dimethicone and trimethylsiloxaysilicate 0.73, cetyl dimethicone 0.45, and polymethylsilsesquioxane 5 g.